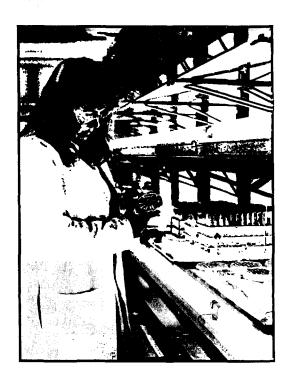


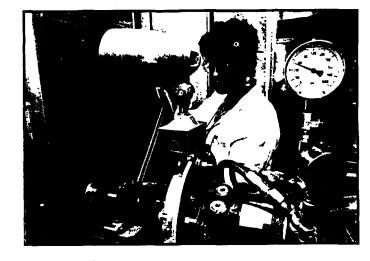
# Bioremediation from the Biofuels People

NREL Has a Wealth of Microorganisms and Technologies to Meet Detoxification Needs

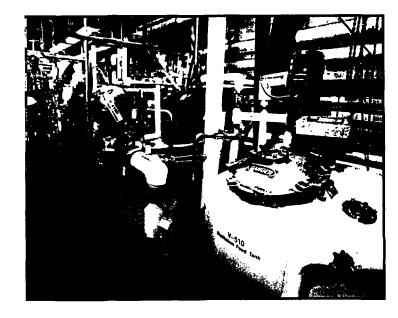
Metals remediation: leachable toxic metal ions are widespread and serious soil, wastewater, and groundwater contaminants. NREL has algae to capture these ions in a slime coating and bacteria that reduce the ions to recoverable metal.



Engineering capabilities: bioremediation effectiveness can be enhanced by innovative reactor designs and optimized systems. NREL's pilot-plant facilities are a key tool for designing the best response to any particular bioremediation need.



Organics destruction: increasingly strict regulations are limiting incineration of these highly toxic waste materials. NREL has anaerobic microbial consortia that convert these compounds to valuable methane and photosynthetic bacteria that degrade them to harmless materials.



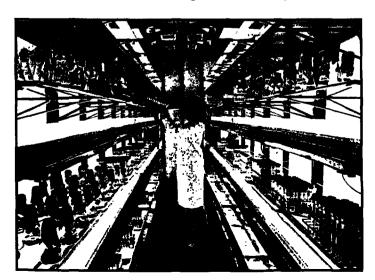




#### Algal Slime Binds Leachable Metals

Many single-cell algae produce a layer of slime around themselves, which works as a selective biological ion exchange resin, because the polysaccharides in the slime layer have an affinity for metal ions in solution. NREL scientist Steve Toon has screened algae for their unique capability to bind metals and—by embedding the algae in a fiberglass matrix in special bioreactors—used them to adsorb metals from acid mine drainage. As contaminated water flows through the reactor, the slime layer traps the metal ions, leaving contaminant-free water to flow on through. Reactor operators periodically harvest the accumulated metals or metal-saturated algae (for disposal or recovery), manipulate the nutrient supply to restore the cell population, change nutrients again to promote slime layer growth, and then put the filters back in service.

The NREL bioremediation team has an extensive microalgae collection to draw upon for metals remediation. The collection was initially developed as part of an effort to use microalgae to capture carbon dioxide and produce lipids (fats and oils) that can be converted to a clean-burning diesel substitute. With their tremendous growth rate, microalgae hold great promise on both counts. NREL researcher Paul Roessler supervises the collection of more than 400 algal strains, many of



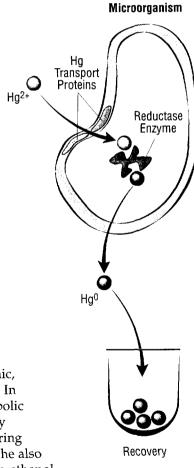
Growth chamber used in screening strains of microalgae for ability to bind metal ions.

which were isolated from unique niches such as saline desert environments.

Some photosynthetic bacteria—one more NREL specialty—also produce polysaccharide slime layers, so they are also biological ion exchange resources. This technology applies to radionuclides and to waste streams of numerous industries such as metal plating, metal finishing, and computer chip manufacturing.

#### **Bacteria Reduce Heavy-Metal Ions**

Another approach to removing metal ion contamination from water or soil is to reduce the metal ions so they are no longer soluble and are therefore easier to concentrate and recover. NREL engineer George Philippidis has expertise for exploiting microorganisms to do this job. He has used the common bacterium Escherichia coli to remediate contamination from soluble mercury ion, a metal contaminant that is one of the most notorious for causing health problems. Bacteria can also reduce a variety of other metals, including lead, copper, arsenic, cadmium, silver, and gold. In this work he applies metabolic modeling, cell-permeability treatment, genetic engineering and other capabilities that he also uses for NREL's biomass-to-ethanol technology program.



Similarly, NREL scientist Ali Mohagheghi has used sulfate-reducing bacteria to recover soluble uranium VI ion. The bacteria produce hydrogen sulfide, which reduces uranium VI ion to form insoluble uranium IV oxide. The bacteria provide a surface for the reaction, and the uranium oxide precipitates out for recovery. The mercury reduction and the uranium precipitation approaches may each solve a variety of heavy metal and radionuclide contamination problems.



## Photosynthetic Bacteria Feed on Creosote, PCB, and Pesticide Contaminants

NREL scientists are developing photosynthetic bacteria for a variety of renewable energy technologies. These bacteria can grow on and produce hydrogen from wastes, biomass, or synthesis gas. In addition, photosynthetic bacteria can produce building blocks for biodegradable plastics. These same bacteria may also be exploited for bioremediation. Recently, Pin-Ching Maness isolated three bacterial strains that can remediate two very important soil and water contaminants. Two of these photosynthetic bacteria digest m-chlorobenzoate—a principal breakdown product of both chlorinated pesticides and polychlorinated biphenyls (PCBs) used in electrical insulators. The other strain digests p-cresol—a principal breakdown product of creosote, a widely used wood preservative. An important advantage of photosynthetic bacteria is that they obtain their energy from the sun, using only the contaminants as a carbon source. This allows them to be effective bioremediation agents in common situations where energy-yielding nutrients necessary for other microorganisms are not readily available.



Photosynthetic bacteria possess unique capabilities to degrade a variety of hazardous organic compounds.



NREL has adapted strict anaerobic microorganisms to degrade a variety of organic wastes to useful by-products including methane.

### Anaerobic Microorganisms Degrade Furfural and Nitriles

One of the many technologies NREL uses to convert municipal solid waste and other biomass wastestreams to energy resources is anaerobic digestion, which generates usable organic compost, as well as methane for power generation. NREL scientist Chris Rivard has adapted natural microbial consortia to degrade furfural. Furfural degradation is a logical objective for NREL because thermochemical pretreatment of biomass for NREL's biomass-to-ethanol fermentation technology produces furfural as an unwanted by-product. Furfural—a chemical that cannot be released without treatment (its uses include killing weeds)—is also generated by sulfite pulping in paper making and during production of fruit juices, wines, and medical solutions.

NREL scientists developed closed reactor systems that increase mixing and capture methane to take maximum advantage of anaerobic digestion. By using these closed systems for bioremediation of organics, they are also preventing these volatile compounds from evaporating. At the same time, the microbial consortium is a totally natural one, so anaerobic digestion can also be safely used in the environment for cleanup of spills.

NREL is now turning its anaerobic digestion methodology sights on a major industrial waste stream—the acetonitrile and cyanide by-products from producing acrylonitrile, a basic chemical that is used to make numerous plastic and rubber materials and is a major U.S. export. Industry now generally incinerates these wastes, a disposal method that is becoming increasingly tightly regulated. Nick Nagle and other NREL scientists are developing anaerobic digestion for bioremediation of acetonitrile and expect that once established it will readily adapt to other nitriles and to cyanide.

# Engineering Capabilities

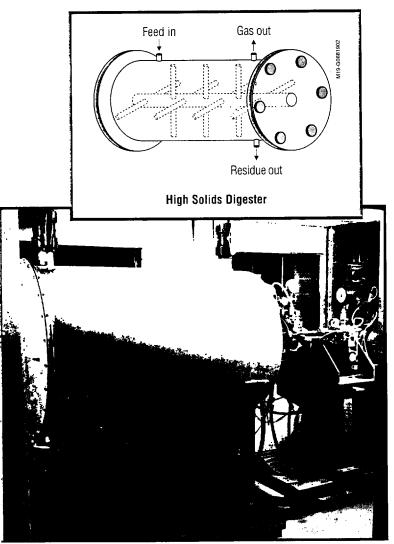
# Reactor Systems and Engineering Capabilities Multiply Bioremediation Effectiveness

In addition to its experience with various microorganisms, a key element of NREL's bioremediation capabilities is the design of innovative reactor systems that maximize the contact between the microorganisms and the waste to be remediated and set optimum contact conditions. For example, the high solids digester Chris Rivard developed for treating mixtures of cellulosic and fatty wastes—a major pilot plant is now being built for municipal solid waste and tuna cannery waste—is far more efficient and economical than the low-solids anaerobic digestion traditionally used in sewage treatment plants. The relatively common practice of "land farming" petroleum refinery or other wastes spreading them on soil for action by the natural mix of microorganisms to work on—could also be more effectively carried out in a reactor such as a highsolids digester. The methane generated would help pay for the cost of the reactor, and contaminants such as volatile organic compounds are totally contained, so they do not "transfer" from a solid waste problem to an air quality problem by evaporation.

The NREL team has a wide range of engineering capabilities used in the development, scale up, and commercialization of biofuels production processes

NREL employs
laboratory scale
fixed film and
upflow anaerobic
sludge blanket
(UASB) systems for
bioremediation of
liquid waste streams
(left) as well as high
solids digesters for
solid wastes (right).

that apply to bioremediation. Team members have expertise in overall system design, computer modeling of the biological and chemical processes, modeling and analysis of project economics, and process integration for understanding the interaction of multiple steps. They also understand how to take advantage of opportunities for recycling and by-product utilization. In addition, NREL's extensive biofuels facilities—including a recently completed biomass-to-ethanol pilot plant—could also be used for bioremediation projects.



#### Putting Expertise to Work

Bioremediation is not simply a matter of throwing a few bugs at a toxic material problem. Effective systems require careful science and effective engineering. NREL has the biofuels resources and experience and the bioremediation expertise to do the job right and is actively seeking industrial partners to forge new advances in bioremediation.

# **Team Members**

#### **Pin-Ching Maness**

Staff Microbiologist, Research and Development B.S. Biology, National Taiwan University (Taipei, Taiwan); M.S. Biochemistry, Indiana State University (Terre Haute, IN) Areas of expertise include bioremediation, photosynthesis, microbial physiology/ biochemistry, and bacterial production of alternative fuels and biodegradable plastics (PHB polymer).

Member: American Society for Microbiology.



#### Ali Mohagheghi

Staff Biochemical Engineer Research and Development

B.S. Chemical Engineering, Sharif University of Technology (Tehran, Iran); M.S. Biochemical Engineering, M.I.T. (Cambridge, MA); Ph.D. Applied Chemistry and Geomicrobiology, Colorado School of Mines (Golden, CO)

Areas of expertise include biochemical engineering, bioremediation, fermentation and enzyme technology, analytical chemistry, geomicrobiology, and geochemistry.

Member: American Society of Microbiology and editorial board for Journal of Letters in Applied Microbiology.

#### Nick Nagle

Environmental Scientist, Research and Development B.S. Microbiology, Colorado State University (Fort Collins, CO); M.S. Environmental Engineering, Colorado School of Mines (Golden, CO)

Areas of expertise include anaerobic digestion, bioreactor system design, bioremediation, biodiesel synthesis from microalgal lipids, and sewage sludge pretreatment.

Member: American Society of Microbiology and Colorado Hazardous Waste Management Society.

#### George Philippidis

Senior Biochemical Engineer, Research and Development Team Leader **B.S. Chemical Engineering**, Aristotle University of Thessaloniki (Thessaloniki, Macedonia, Greece); Ph.D. Chemical Engineering, University of Minnesota (Minneapolis, MN); M.B.A., University of Denver (Denver, CO)

Areas of expertise include biochemical engineering, bioremediation, fermentation and enzyme technology, genetic engineering, mathematical modeling, and project management Member: American Institute of Chemical Engineers and American Management Association

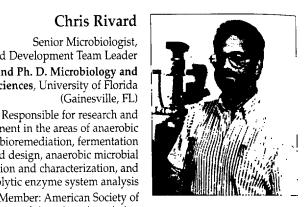


#### Chris Rivard

Senior Microbiologist, Research and Development Team Leader B.S., M.S., and Ph. D. Microbiology and Cell Sciences, University of Florida (Gainesville, FL)

development in the areas of anaerobic digestion, bioremediation, fermentation systems and design, anaerobic microbial isolation and characterization, and hydrolytic enzyme system analysis Member: American Society of Microbiology and American Association

for the Advancement of Science



#### Paul Roessler

Senior Biologist

Reasearch and Development Team Leader B.S. Biology, Grove City College (Grove City, PA); M.S. Botany, Michigan State University (East Lansing, MI); Ph.D. Biology, University of Colorado (Boulder, CO)

Areas of expertise include plant/algal biochemistry and molecular biology, protein purification, gene cloning, algal genetic engineering, lipid analysis, microalgal screening

Member: Phycological Society of America, American Society of Plant Physiologists, American Oil Chemists' Society

#### Steve Toon

Associate Scientist, Research and Development B.S. Microbiology, M.S. Botany, University of Arkansas (Fayetteville, AR); Ph.D. candidate Biological Sciences, University of Denver (Denver, CO)

Areas of expertise include microbiology, biochemistry, photobiology, bioreactor design and operation, microbial prospecting, bioremediation, industrial strain development, and photocatalytic sterilization Member: American Society of

Plant Physiology





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